per second, at least about 500,000 droplets per second, at least about 750,000 droplets per second, at least about 1,000,000 droplets per second, at least about 1,500,000 droplets per second, at least about 2,000,000 or more droplets per second, or at least about 3,000,000 or more droplets per second may be produced.

[0065] In some aspects, the fluidic droplets may also contain additional entities, for example, other chemical, biochemical, or biological entities (which may be dissolved or suspended in the fluid in some cases), for example, monomers, polymers, metals, magnetizable materials, cells, beads, gases, other fluids, or the like. Examples of entities or species that may be contained within, or otherwise associated with, a fluidic droplet include, but are not limited to, signaling entities such as those described below, pharmaceutical agents, drugs, hormones, nucleic acids such as DNA or RNA, proteins (e.g., antibodies), peptides, fragrance, reactive agents, biocides, fungicides, preservatives, chemicals, cells, and the like, as well as combinations thereof. For example, a droplet may contain an antibody-producing cell and an entity which the antibodies produced by the cell can interact with, such as another cell, an antigen, a protein, or the like. Such entities may be useful, for example, in an assay to determine the antibody within the droplet.

[0066] Numerous other cell-based assays are possible, including those that monitor cell response to stimuli. For example, cells can be encapsulated with drugs from a drug compound library and assayed for cell death. Additionally or alternatively, target cells can be genetically modified so that a desired antibody binding to a cell surface protein transmits a signal resulting from cellular production of a signaling entity, e.g., green fluorescent protein. These "read-out" cells can be encapsulated with a library of antibody-secreting cells and cells that produce the desired antibody can be isolated and identified.

[0067] Thus, in one aspect, a characteristic of a droplet is determined in some fashion, e.g., to determine a species contained within a fluidic droplet. For instance, a species such as a protein, a polypeptide, a peptide, a nucleic acid, an antibody, an enzyme, a virus, a hormone, or the like is determined within the fluidic droplet, and in some cases, the fluidic droplet is processed in some fashion as a result of that determination (e.g., screened and/or sorted, as discussed below).

[0068] In one set of embodiments, a signaling entity may be used to determine the characteristic. For instance, a signaling entity may be present within the fluidic droplet and/or within the liquid surrounding the fluidic droplet. Examples of characteristics that may be determined by the signaling entity include, but are not limited to, the presence or concentration of a species, the activity of the species (e.g., the binding activity, catalytic activity, regulatory activity, etc.), and the relative activity of one species compared to another species, etc. In some cases, more than one signaling entity may be used, and in some cases, two or more different, distinguishable signaling entities may be used, e.g., signaling entities able to bind the same or different species. In some embodiments, one or more signaling entities may facilitate the determination of an entity's ability to generate a particular species inside the fluidic droplet (e.g., determination of a cell's ability to produce a particular antibody). In yet other embodiments, one or more signaling entities may facilitate the determination of an entity's response to a particular species (e.g., the response of a cell to a toxin).

[0069] As used herein, a "signaling entity" means an entity that is capable of indicating its existence in a particular sample or at a particular location. Signaling entities of the invention can be those that are identifiable by the unaided human eye, those that may be invisible in isolation but may be detectable by the unaided human eye if in sufficient quantity (e.g., microparticles), entities that absorb or emit electromagnetic radiation at a level or within a wavelength range such that they can be readily detected visibly (unaided or with a microscope including an electron microscope or the like), or spectroscopically, or the like. Examples include dyes, pigments, fluorescent moieties (including, by definition, phosphorescent moieties), up-regulating phosphors, chemiluminescent entities, electrochemiluminescent entities, or enzymatic signaling moieties including horseradish peroxidase and alkaline phosphatase.

[0070] In one set of embodiments, a signaling entity may comprise a microparticle and an agent immobilized relative to the microparticle that is able to bind, specifically or non-specifically, to a species to be determined, for example, as a protein, a polypeptide, a peptide, a nucleic acid, an antibody, an enzyme, a hormone, or the like. The agent may be immobilized to the microparticle covalently or non-covalently. The agent may be immobilized directly to the microparticle or via a linker. The microparticles typically will have an average diameter (defined as above) of less than about 1 mm, and can be spherical or non-spherical.

[0071] In one set of embodiments, the agent is a binding partner of the species to be determined. A "binding partner," as used herein, refers to any molecule that can undergo binding with a particular molecule. For example, Protein A is a binding partner of the biological molecule IgG, and vice versa. Other non-limiting examples include nucleic acid-nucleic acid binding, nucleic acid-protein binding, protein-protein binding, enzyme-substrate binding, receptor-ligand binding, receptor-hormone binding, antibody-antigen binding, etc. Binding partners include specific, semi-specific, and non-specific binding partners as known to those of ordinary skill in the art. For example, Protein A is usually regarded as a "non-specific" or semi-specific binder.

[0072] The term "specifically binds," when referring to a binding partner (e.g., protein, nucleic acid, antibody, etc.), refers to a reaction that is determinative of the presence and/or identity of one or other member of the binding pair in a mixture of heterogeneous molecules (e.g., proteins and other biologics). Thus, for example, in the case of a receptor/ligand binding pair the ligand would specifically and/or preferentially select its receptor from a complex mixture of molecules, or vice versa. An enzyme would specifically bind to its substrate, a nucleic acid would specifically bind to its complement, an antibody would specifically bind to its antigen. Other examples include nucleic acids that specifically bind (hybridize) to their complement, antibodies specifically bind to their antigen, binding pairs such as those described above, and the like. The binding may be by one or more of a variety of mechanisms including, but not limited to ionic interactions, and/or covalent interactions, and/or hydrophobic interactions, and/or van der Waals interactions, etc.

[0073] In one set of embodiments, a first signaling entity may be allowed to bind the species to be determined, and a second signaling entity allowed to bind the first entity. One or both of the first or second signaling entities may be determinable, e.g., fluorescent. Higher-order determinations are also contemplated. For instance, a first signaling entity may be